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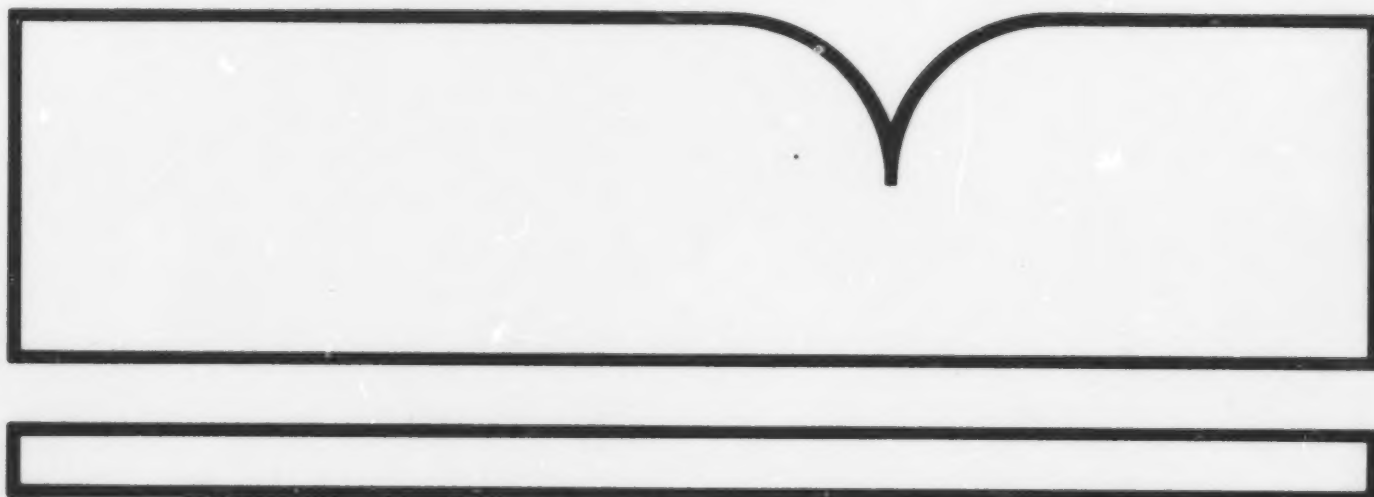
Multicompartment Kinetic Model for Lead
Part 3. Lead in Blood Plasma and
Erythrocytes

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Multicompartment Kinetic Model for Lead

III. Lead in Blood Plasma and Erythrocytes

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Multicompartment models have been fitted to experimental data on plasma lead and blood lead concentrations of subjects studied by P. E. deSilva (*Brit. J. Ind. Med.* 38, 209-217, 1981) and one subject studied by W. I. Manton and C. R. Malloy (*Brit. J. Ind. Med.* 40, 51-57, 1983). Nonlinear models for plasma lead vs blood lead in populations provide significantly better descriptions than does a linear model. Short-term kinetic data do not clearly resolve the precise nonlinear mechanism, however. Parameters of plasma and erythrocyte distribution in a single subject were significantly different on two occasions. © 1985 Academic Press, Inc.

1. INTRODUCTION

There is little doubt that at high levels of lead exposure, blood lead in humans is governed by some nonlinear mechanism or mechanisms. Much of the evidence for the existence of nonlinear kinetics rests on nonlinearity of the "dose-response" relationship between blood lead and lead concentrations in environmental media to which the subjects are exposed. The blood lead vs. air lead relation has been reviewed by a number of authors (Hammond *et al.*, 1981; Snee, 1982; Chamberlain, 1983; Laxen, 1983; Brunekreef, 1984.) using essentially the same set of observational and experimental studies. Chamberlain (1983) has concluded that after several months of steady exposure to environmental lead, blood lead levels achieve a near-equilibrium concentration that increases linearly with the ambient concentration no matter what the exposure pathway (directly by air inhalation, or by ingestion of food, water, dust, soil, or paint), provided the total exposure does not cause blood lead to exceed 30 to 40 $\mu\text{g/dl}$. However, when total lead exposure by any pathway becomes so great that blood lead levels greatly exceed 60 to 80 $\mu\text{g/dl}$, then the blood lead concentrations increase much more slowly with increasing exposure concentration than they do at lower levels.

We have investigated some of the possible mechanisms for this nonlinearity, based on the kinetic response of blood lead to changes in body lead burden (Marcus, 1983, 1985a, b). None of the nonlinear mechanisms tested by our multicompartment kinetic analyses provided a significantly improved fit to experimental data at normal blood levels, except possibly for a change in the rate of lead absorption from the gastrointestinal tract that may be time dependent, but was not dose dependent. In this note we will statistically analyze the separate components of the blood lead pool, particularly plasma lead, since it is believed

that plasma lead is the most toxicologically active component (deSilva, 1981a, b). Strong evidence exists for nonlinear kinetics, and statistical analyses of some recent data sets can identify the most plausible of several models proposed for these data.

The underlying problem is that it is very difficult to measure lead concentrations in blood plasma. All of the more recent papers (e.g., Cavalleri *et al.*, 1978; Everson and Patterson, 1980; deSilva, 1981a, b) have developed improved methods for plasma lead measurement and have criticized earlier studies. We have thus used the most recent data of deSilva (1981a) and Manton and Malloy (1983) for our analyses.

2. DATA USED IN THE ANALYSES

deSilva's data (1981) included both a large sample of lead workers with blood lead levels high enough to show nonlinear processes, and two short-term experiments on the same subjects to allow an independent determination of the kinetic model. The data on hematocrit, plasma, and blood lead concentrations in 105 Australian workers were kindly provided by Ms. deSilva. The subjects were selected, to some extent, on the basis of their blood lead levels and on related criteria such as screening coproporphyrin test. Thus the study sample cannot be construed as representative of some population. However, these data are quite adequate for establishing a regression relationship that would allow prediction of the relatively less precise plasma lead levels from the relatively more precise blood lead levels. Duplicate measurements were available for 88 of the plasma leads, yielding 193 samples. Two subjects had duplicate plasma leads that differed by at least a factor of two; omitting these two yielded 189 samples on 103 subjects. Three of the subjects remaining had hematocrits less than 36%; omitting these three left 100 subjects with 183 samples.

A single subject ingested 100 mg of lead as lead acetate on two occasions, in August and December 1979. We need hardly emphasize the uniqueness of this experiment, which involved rather high maximum blood lead levels (78 $\mu\text{g}/\text{dl}$ in August and 46 $\mu\text{g}/\text{dl}$ in December). Nine blood samples were taken at various times during each experiment.

An additional single-subject data set has been presented in (Manton and Malloy, 1983). A 60-year-old male exposed to lead solder (possibly ingested by choice) had elevated levels of blood and plasma-lead. Eight samples were taken at least 3 days apart. While the unknown pattern of lead intake and removal by chelation therapy does not allow meaningful kinetic modelling, the overall serum-blood curve is quite similar in shape to that of deSilva's workers and was analyzed similarly.

3. KINETIC MODELS FOR PLASMA LEAD VS BLOOD LEAD IN POPULATIONS

Cross-sectional observational data from lead-exposed populations do not allow direct inferences on nonlinear kinetic mechanisms of lead distribution within the body and elimination from the body. However, useful indirect inferences may be made when measurements from each subject include the concentration of plasma

1, denoted $[P]$, and the concentration of lead in blood, $[B]$. It is here convenient to define other quantities in the model:

- P = mass of lead in plasma, μg
- B = mass of lead in whole blood, μg
- E = mass of lead in erythrocytes, $\mu\text{g} = B - P$
- t = time, days
- V_P = plasma volume, dl
- V_B = blood volume, dl
- K_{EP} = fractional rate at which lead in plasma is transferred to erythrocytes, per day
- K_E = fractional rate at which lead is lost from erythrocytes to plasma, per day
- Ht = hematocrit = $1 - V_P/V_B$
- $[P]$ = plasma lead concentration = P/V_P , $\mu\text{g}/\text{dl}$
- $[B]$ = blood lead concentration = B/V_B , $\mu\text{g}/\text{dl}$
- $R = K_{EP}/K_E$ = ratio of lead flowing into and out of erythrocytes
- M = mass at which some nonlinear rate parameter changes by 100% (M_E or M_P)
- $[M]_P = M_P/V_P$, $\mu\text{g}/\text{dl}$ in plasma
- $[M]_B = M_E/V_B$, $\mu\text{g}/\text{dl}$ in blood.

It is convenient to assume that the mass of lead in various coupled organs or physiological systems evolves as system of coupled first-order differential equations. The equilibrium relationship between plasma and blood lead depends only on the equation in which erythrocyte and plasma pools exchange lead. We here show these equilibrium relationships for five plausible models.

Model 0. Linear kinetics:

$$dE/dt = K_{EP}P - K_E E. \quad (1)$$

Setting $dE/dt = 0$ we obtain from (1):

$$\begin{aligned} E &= RP \\ B &= (1 + R)P \text{ or} \\ [P] &= [B]/(1 + Ht(1 + R)). \end{aligned} \quad (2)$$

Model 1. Limited plasma flow into erythrocytes (1A), or accelerated flow into plasma (1B) as plasma lead increases:

$$(1A) \quad dE/dt = K_{EP}P(1 - P/M) - K_E E \quad (0 \leq P \leq M) \quad (3)$$

$$(1B) \quad dE/dt = K_{EP}P - K_E E/(1 - P/M) \quad (0 \leq P \leq M). \quad (4)$$

These are analyzed together, since $dE/dt = 0$ yields for both (1A) and (1B)

$$[P] = \{R + 1 - ((R + 1)^2 - 4R[B]/[M]_P(1 - Ht))^{0.5}\}/[M]_P/2R \quad (5)$$

which cannot be distinguished by cross-sectional data alone.

Model 2. Decreased rate of flow lead from plasma into erythrocytes as plasma lead increases (2A), or linearly accelerated flow into plasma (2B) as plasma lead increases:

$$(2A) \quad dE/dt = K_{EP}P/(1 + P/M) - K_E E \quad (0 \leq E \leq RM) \quad (6)$$

$$(2B) \quad dE/dt = K_{EP}P - K_E E(1 + P/M) \quad (0 \leq E \leq RM). \quad (7)$$

Setting $dE/dt = 0$, these both yield

$$[P] = \{(C_2^2 + 4[B]/[M]_p(1 - Ht))^{0.5} - C_2\}[M]_p/2 \quad (8)$$

where

$$C_2 = R + 1 - [B]/[M]_p(1 - Ht).$$

Model 3. Decreased rate of flow of lead from plasma into erythrocyte as erythrocyte lead increases (3A), or linearly accelerated flow into plasma (3B) as erythrocyte lead increases:

$$(3A) \quad dE/dt = K_{EP}P/(1 + E/M) - K_E E \quad (9)$$

$$(3B) \quad dE/dt = K_{EP}P - K_E E(1 + E/M). \quad (10)$$

Setting $dE/dt = 0$, these both yield

$$[P] = \{C_3 - (C_3^2 - 4[B]([B] + [M]_B)^{0.5})/[M]_B(1 - Ht)\} \quad (11)$$

where

$$C_3 = R + 1 + 2[B]/[M]_B.$$

Model 4. Limited flow of lead from plasma into erythrocyte as erythrocyte lead increases (4A), or nonlinearly accelerated rate of flow into plasma as erythrocyte lead increase (4B):

$$(4A) \quad dE/dt = K_{EP}P(1 - E/M) - K_E E \quad (0 \leq E \leq M) \quad (12)$$

$$(4B) \quad dE/dt = K_{EP}P - K_E E/(1 - E/M) \quad (0 \leq E < M). \quad (13)$$

Setting $dE/dt = 0$ yields

$$[P] = \{-C_4 + (C_4^2 + 4R[B]/[M]_B)^{0.5}\}[M]_B/2R(1 - Ht) \quad (14)$$

where

$$C_4 = R + 1 - R[B]/[M]_B.$$

The five distinguishable models, given by Eqs. (2), (5), (8), (11), and (14), can be fitted to data sets such as those in (deSilva, 1981a, b) in which individual measurements of $[B]$, $[P]$ and Ht are available for each subject. All of these models have an unknown kinetic parameter R that can be estimated from the data using standard nonlinear regression procedures. We used the nonlinear regression procedure PROC NLIN in the Statistical Analysis System (SAS) (1982). Models 1-4 also have an unknown parameters $[M]$ that gives the concentration of lead in plasma or blood at which the nonlinear lead transfer rate changes by 100% from its value at 0. For Models 1 and 4 this is the value at which the lead-holding capacity, in plasma and erythrocytes, respectively, is completely saturated. Note that with *constant* hematocrit assumed, Models 2 and 4 (Eqs. (8) and (14)) give statistically equivalent results.

Models 0-4 were fitted to deSilva's worker data with one or two observations per worker, samples sizes of 193 (all cases), 189 (omitting two subjects), and 183 (omitting five subjects). Residual variances were smallest with only two subjects omitted, $N = 189$, and all analyses are reported on this basis. In order to account for the greater variability at larger plasma lead concentrations, (see Fig. 1) weighted as well as unweighted estimates were calculated. The results were not

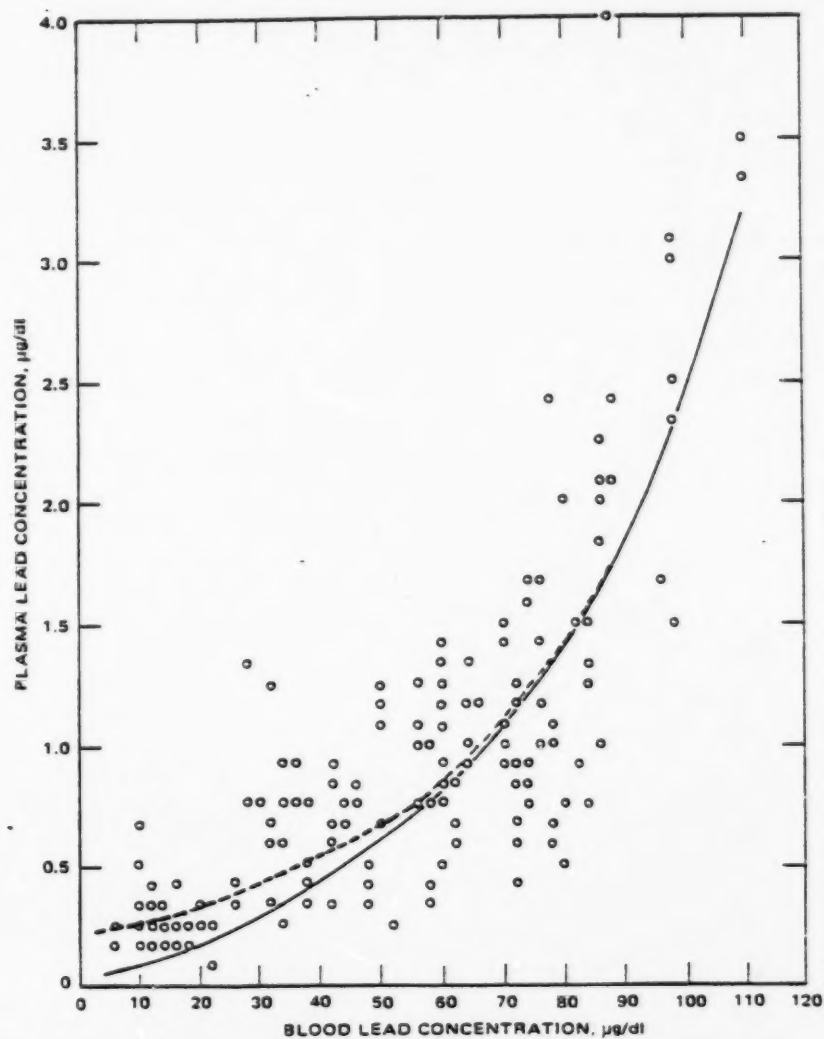


FIG. 1. Nonlinear kinetic Model 4 fitted to data from 189 samples from 103 workers, as reported in deSilva (1981a, b). Solid curve is no-intercept model, and dashed curve is model with intercept = 0.24 $\mu\text{g/dl}$.

changed much by the weighting method, and only unweighted estimates are reported. All models were also fitted with an intercept parameter to allow for systematic bias in plasma measurements, possibly attributable to contamination. This intercept term was significant in most models and estimates in Tables 1-3 are shown both with and without an intercept term in Table 4. The fitted curve for Models 2 or 4, unweighted, with constant hematocrit and no intercept, is shown in Fig. 1 to emphasize the utility of an intercept term.

In all cases, reliable estimates were obtained for the nonlinear concentration parameter $[M]$. In all cases, the nonlinear models provided a significantly better fit to the data than did the corresponding linear Model 0, using an asymptotically valid statistical F test (Gallant, 1975). For Models 1 and 4, all observed $[P] < [M]_p$ and observed $[B] < [M]_b$. The large estimated standard errors on some $[M]$ suggest that they are not precisely determined, and should not be used for confidence interval estimates. For Models 2 and 4 the intercept term allowed a significantly better fit to the data, suggesting a nonnegligible displacement of measured plasma lead concentrations.

Other interesting hypotheses could not be formally tested in this manner, since the hypotheses were not nested within each other, i.e., were not hierarchical. Nonetheless, some informal comparisons are very instructive. Contrary to our expectations, the model with constant hematocrit = 0.45 always provided a better fit to the data than did the model with individual hematocrit substituted for Ht. For example, for Model 4, the relative improvement in residual sum of squares,

TABLE I
RESIDUAL SUMS OF SQUARES (SSE) FOR NONLINEAR MODELS
(RESIDUAL MEAN SQUARES IN PARENTHESES)

Intercept term:	Hematocrit			
	Individual		All hematocrits = 0.45	
	No	Yes	No	Yes
Model 0	40.735 (0.2167)	40.632 (0.2161)	37.406 (0.1990)	37.026 (0.1969)
1	36.444 (0.1949)	36.297 (0.1951)	30.181 (0.1614)	33.813 (0.1818)
2	36.368 (0.1945)	33.860 (0.1820)	29.381 (0.1571)	26.890 (0.1446)
3	35.733 (0.1911)	34.780 (0.1870)	32.621 (0.1744)	33.611 (0.1807)
4	31.703 (0.1695)	28.202 (0.1516)	29.381 (0.1571)	26.890 (0.1446)
Parameters, P for Models 1-4	2	3	2	3
N	189			
Total SS	83.154			

TABLE 2
NONLINEAR CONCENTRATION PARAMETER $[M]$ $\pm$ STANDARD ERROR ($\mu\text{g/dl}$) IN PLASMA OR BLOOD

Intercept term:	Hematocrit			
	Individual		All Hematocrits = 0.45	
	No	Yes	No	Yes
Plasma				
Model 1	5.34 $\pm$ 0.37	5.14 $\pm$ 0.25	5.59 $\pm$ 0.18	5.39 $\pm$ 0.69
Model 2	1.49 $\pm$ 0.28	0.68 $\pm$ 0.15	1.27 $\pm$ 0.15	0.72 $\pm$ 0.11
Blood				
Model 3	35.8 $\pm$ 21.5	14.1 $\pm$ 11.7	40.8 $\pm$ 22.0	51.5 $\pm$ 94.3
Model 4	142.6 $\pm$ 6.3	126.2 $\pm$ 3.7	149.7 $\pm$ 7.6	130.9 $\pm$ 4.7

SSE, defined as $(\text{SSE}(\text{individual}) - \text{SSE}(\text{constant Ht})) / (\text{SSE}(\text{constant Ht}) / (N - P))$, is $(31.703 - 29.381) / (0.1571) = 14.78$ for the no-intercept models, and 9.07 for the intercept model. The null hypothesis sampling distribution of this criterion for small samples is not known, but by analogy with the F distribution it appears large. (See Pereira (1977) for a bibliography on related problems.) That is, the assumption of a constant hematocrit in the population actually improves the fitted model even though the use of individual hematocrit values is physiologically realistic.

We note finally that for the models with individual hematocrit, Model 4, is much better than any of the others. For constant hematocrit, Models 2 and 4 give equivalent fit to the data, and they are very much better than the others. In order to distinguish between them, it is necessary to examine in detail experimental data on blood lead kinetics in humans, which we do in the next section.

The estimates of $[M]$ are given in Table 2, estimates of R in Table 3, and estimates of the intercept in Table 4. It is seen that the choice of individual or constant hematocrit little affects the Model 0 or 1 estimates, but strongly affects the Model 2, 3, and 4 estimates. The use of an intercept is highly correlated with $[M]$ and R estimates. However, the intercept corresponds to a plasma concentration of about 0.25–0.30 $\mu\text{g/dl}$ in Models 2, 3, and 4, which is significantly greater

TABLE 3
KINETIC TRANSFER RATIO R (INTO ERYTHROCYTES/OUT OF ERYTHROCYTES) $\pm$ STANDARD ERROR FOR LINEAR AND NONLINEAR MODELS

Intercept term:	Hematocrit			
	Individual		All Hematocrits = 0.45	
	No	Yes	No	Yes
Model 0	108.0 $\pm$ 3.7	103.2 $\pm$ 3.3	104.6 $\pm$ 3.4	96.0 $\pm$ 2.8
1	143.0 $\pm$ 6.0	149.2 $\pm$ 215.9	139.9 $\pm$ 4.4	145.1 $\pm$ 13.5
2	202.2 $\pm$ 19.8	350.3 $\pm$ 59.1	214.5 $\pm$ 15.1	328.3 $\pm$ 39.3
3	325.4 $\pm$ 132.8	885.8 $\pm$ 2031.7	289.8 $\pm$ 101.6	310.9 $\pm$ 294.3
4	234.1 $\pm$ 16.5	373.4 $\pm$ 43.4	214.5 $\pm$ 15.1	328.3 $\pm$ 39.3

TABLE 4
INTERCEPT TERM (PLASMA AT BLOOD = 0) $\pm$ STANDARD ERROR FOR
LINEAR AND NONLINEAR MODELS

Model	Hematocrit	
	Individual	All Hematocrits = 0.45
0	-0.05 $\pm$ 0.05	-0.10 $\pm$ 0.05
1	0.05 $\pm$ 0.06	0.20 $\pm$ 0.07
2	0.26 $\pm$ 0.06	0.24 $\pm$ 0.06
3	0.31 $\pm$ 0.10	0.25 $\pm$ 0.11
4	0.27 $\pm$ 0.05	0.24 $\pm$ 0.06

than 0 in Models 2, 3, and 4. Since the intercept improves these fitted model by a large amount, we recommend using it.

deSilva also called attention to the possibility that four individuals with high ratios of plasma lead to erythrocyte lead may not have achieved plasma equilibrium due to very recent lead exposure. We eliminated 5 samples with $[P] > 2.6$ $\mu\text{g/dl}$ and repeated the analysis as before. The results still showed a highly significant departure from linearity. For the model with an intercept term and $Ht = 0.45$, the comparison of Models 2 and 4 vs Model 0 yielded $F(1,181) = 17.44$, a highly significant improvement. One would have to eliminate most of the data with $[B] > 80$ $\mu\text{g/dl}$ to eliminate the nonlinearity.

An additional small data set of this type in (Manton and Malloy, 1983) was analyzed in the same way. The intercept term was not significant and the fitted models (assuming $Ht = 0.45$) are shown in Table 5. Models 2-4 give a much better fit to the data than Model 0 but the improvement in SSE ($F(1,5) = 1.91$ for Models 2 and 4, $F(1,5) = 3.08$ for Model 3) is not statistically significant at the usual 5% significance level.

4. COMPARTMENTAL MODELS FOR SHORT-TERM PLASMA AND ERYTHROCYTE KINETICS

It is clear from Table 2, Models 2 and 4, that significant nonlinear effects in plasma and total blood lead are likely only going to be detected at high levels of blood or plasma lead concentration. Genuine experiments at such high levels are unusual, and those reported by deSilva (1981a, b) are most suitable for nonlinear kinetic analysis using the techniques of compartmental analysis (Jacquez, 1972).

TABLE 5
LINEAR AND NONLINEAR KINETIC PARAMETERS FOR THE SUBJECT IN (MANTON AND MALLOY, 1983)

Model	SSE	MSE	$[M]$	R
0	1.384	0.1977	—	127.0 $\pm$ 20.2
1	1.135	0.1892	5.52 $\pm$ 3.15 Plasma	165.7 $\pm$ 45.0
2	1.050	0.1750	2.08 $\pm$ 1.91 Plasma	203.1 $\pm$ 80.2
3	0.914	0.1524	20.9 $\pm$ 61.1 Blood	634.0 $\pm$ 1509
4	1.050	0.1750	233.2 $\pm$ 122.5 Blood	202.5 $\pm$ 80.2
Total SS	3.825			

The assumed compartment model is shown in Fig. 2. We have also assumed the following: (i) absorption from gut is a monoexponential process with a constant fraction of the initial dose absorbed into the blood; (ii) lead moves quickly from blood plasma (compartment 2) to the "shallow" pool of the red blood cells (compartment 3), and then much more slowly into the "peripheral" pool of the red blood cells. Lead is held in the peripheral compartment 4 for a relatively long time. Certain experiments suggest that such a model may be a reasonable description. The uptake of lead by human erythrocytes *in vitro* was studied by Clarkson and Kench (1958) who suggested that lead may form a "precipitate or larger particulate form of lead phosphate" on the surface of the blood cells, which is slowly dispersed and is removed only slowly by the usual chelating agents. In a report by Stover (1959) a very similar 4-compartment model is sketched on p. 271. In a subsequent study (Bruenger *et al.*, 1973) on the association of lead with various components of red blood cells, it is shown that about 10% of ^{210}Pb was associated with the stroma and 90% with the cytosol. Thus, without specifying in detail the nature of the two lead compartments in the red blood cells, it appears quite useful to include them in compartmental model to the extent that the data support reliable estimates of their parameters. The parameters for lead kinetics in blood can be estimated reasonably well from the crude compartmental model developed here, except for the exchange with the peripheral soft tissue compartments, which are relatively long lived (Rabinowitz *et al.*, 1976; Marcus, 1983; 1985a, b) and so cannot be reliably estimated from 2 to 9 days of data. The basic model is described in Appendix 1.

deSilva noted that the hematocrit (fraction of blood volume in erythrocytes) varied during the experiment and that a correction for hematocrit appears to be necessary. The hematocrit values covered only a small normal range of values, 46 to 49% during the 9 days of the August experiment, and 46 to 48% during the 2 days of the December experiment. No systematic pattern of variation was

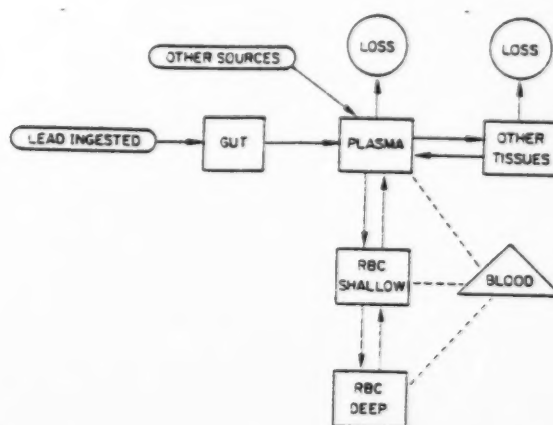


FIG. 2. Compartmental model for lead in plasma and red blood cells.

apparent. The Ht data were fitted as cubic functions of time and used to adjust plasma or erythrocyte volumes respectively, assuming the other held constant.

No significant changes resulted, although constant plasma volume improved the total blood lead model slightly and constant erythrocyte volume improved the plasma lead model slightly. All analyses are reported here for constant hematocrit set equal to the initial estimated value (0.4739 for August, 0.475 for December).

The nonlinear kinetic parameters were estimated with reasonable precision and are exhibited in Table 6. The observed and fitted quantities of blood lead are shown in Figs. 3 and 4 for the August and December experiments, and plasma lead in Figs. 5 and 6, assuming a linear kinetic model. In view of the substantial experimental difficulties in measuring plasma lead concentrations, and the unincorporated random variations such as hematocrit, the agreement between model and data is quite reasonable. As usual, we cannot claim that the fitted model has been correctly identified, and there are other models that would have given the same results since some peripheral compartments cannot be observed. We have however shown that an adequate description of the data can be obtained with the usual structure shown in Fig. 2. The specification of the nonlinear models is also shown in Table 6.

These analyses provided only a limited degree of resolution of the nonlinear kinetic mechanism. Due to the small number of data points and large number of adjustable parameters, none of the differences are statistically significant. As seen in Table 6, the "B" version of Models 1-4 (accelerated flow from shallow erythrocyte pool into plasma) are inferior to the "A" version (reduced rate of flow from plasma to erythrocyte). Likewise, the maximum observed plasma and blood lead concentrations, which occurred 2 hr after ingesting the lead acetate solution, were predicted about as well or better by the "A" version. Combining these criteria, the most acceptable nonlinear models are 1A, 2A, and 3A. However, the value of $[M]$ for Model 1A is so large that this model is, in effect, virtually linear.

Five additional nonlinear models specifying linear blood kinetics were also fitted to the August data. None of these provided quite as good description of the data as the above models.

It is noteworthy—one may even say striking—that the kinetic parameters for the same individual differed so greatly between the August and December experiments, as shown in Table 7. This was of course noted by deSilva, who stated that "This may have been due to the fact that the subject's fluid intake was much higher in the second study and slowed down the absorption of lead from the gut." The effects of environmental factors on internal kinetics is clearly important here. The long-term parameters are not as different between the two experiments as are the short-term parameters. More work needs to be done on the development of predictive models for the partitioning of blood lead among plasma and cells.

5. DISCUSSION

A nonlinear relationship exists between plasma lead and blood lead in deSilva's study (1981a, b). deSilva attributes some of the large plasma lead to erythrocyte lead ratios to "departures from the equilibrium condition, and such departures

TABLE 6
NONLINEAR COMPARTMENTAL MODELS FOR BLOOD LEAD^a

Model	Nonlinear fractional transfer rate	Weighted residual SSE	(Predicted maximum) (Observed maximum)		Nonlinear parameter [M] ± SE
			Plasma	Blood	
0. Linear kinetics	—	134.2	1.011	0.891	—
1A. Plasma site limited flow, to RBC	$k(3,2)/(1 - X(2)/M)$	128.8	1.010	0.903	95 ± 52 µg/dl plasma
1B. Plasma site accelerated flow, to plasma	$k(2,3)/(1 - X(2)/M)$	137.6	0.951	0.895	54 ± 48 µg/dl plasma
2A. Plasma rate limited flow, to RBC	$k(3,2)/(1 + X(2)/M)$	129.3	1.016	0.906	29 ± 21 µg/dl plasma
2B. Plasma rate accelerated flow to plasma	$k(2,3)/(1 + X(2)/M)$	138.0	0.996	0.903	7.3 ± 4.9 µg/dl plasma
3A. RBC rate limited flow, to RBC	$k(3,2)/(1 + X(3)/M)$	130.3	1.002	0.902	97 ± 85 µg/dl blood
3B. RBC rate accelerated flow to plasma	$k(2,3)/(1 + X(3)/M)$	135.2	0.997	0.895	131 ± 131 µg/dl blood
4A. RBC site limited flow, to RBC	$k(3,2)/(1 - X(3)/M)$	140.8	0.998	0.894	220 ± 187 µg/dl blood
4B. RBC site accelerated flow, to plasma	$k(2,3)/(1 - X(3)/M)$	149.0	1.002	0.885	232 ± 153 µg/dl blood
5. RBC rate limited flow, shallow to deep	$k(4,3)/(1 + X(3)/M)$	141.2	0.999	0.889	59 ± 35 µg/dl blood
6. RBC site limited flow, shallow to deep	$k(4,3)/(1 - X(4)/M)$	137.1	1.010	0.890	146 ± 127 µg/dl blood
7. Increased plasma clearance	$k(0,2)/(1 + X(2)/M)$	138.0	0.986	0.896	2.7 ± 2.4 µg/dl plasma
8. Increased tissue absorption	$k(5,2)/(1 + X(2)/M)$	140.4	1.011	0.881	1.8 ± 1.8 µg/dl plasma
9. Rate-limited gut absorption into plasma	$k(2,1)/(1 + X(1)/M)$	137.6	0.999	0.882	1565 ± 1393 µg gut

^a For blood lead levels reported for deSilva's August experiment (1981a, b).

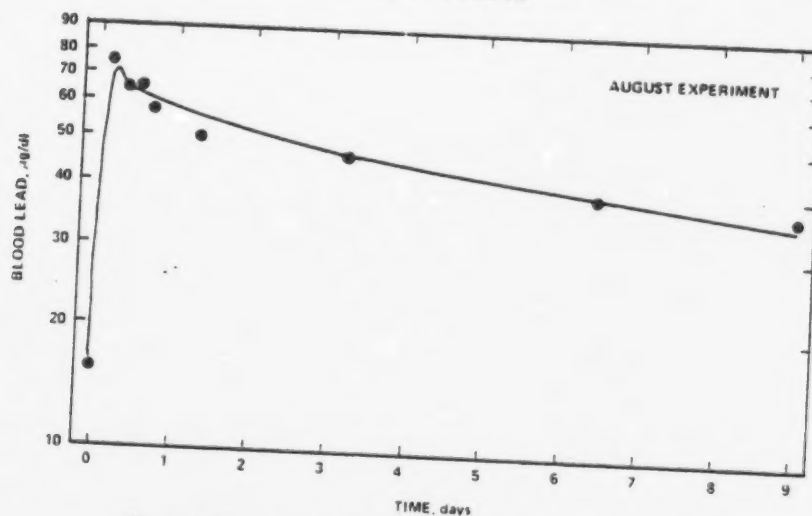


FIG. 3. Observed and predicted blood lead in August experiment.

could be expected to occur immediately after sudden high intakes of lead. . . . Two of the four subjects [circled points in Fig. 2 (deSilva, 1981b)] were maintenance men whose exposure to lead is likely to be high but intermittent. One of

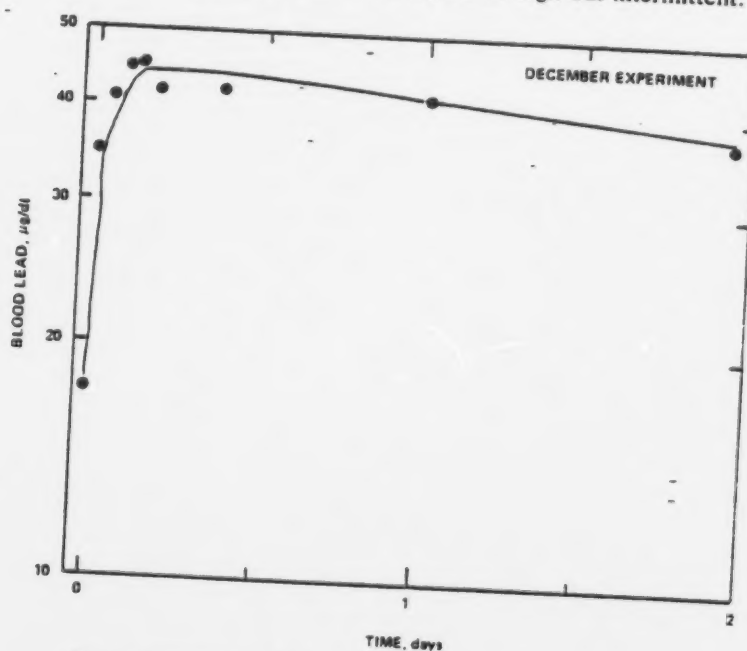


FIG. 4. Observed and predicted blood lead in December experiment.

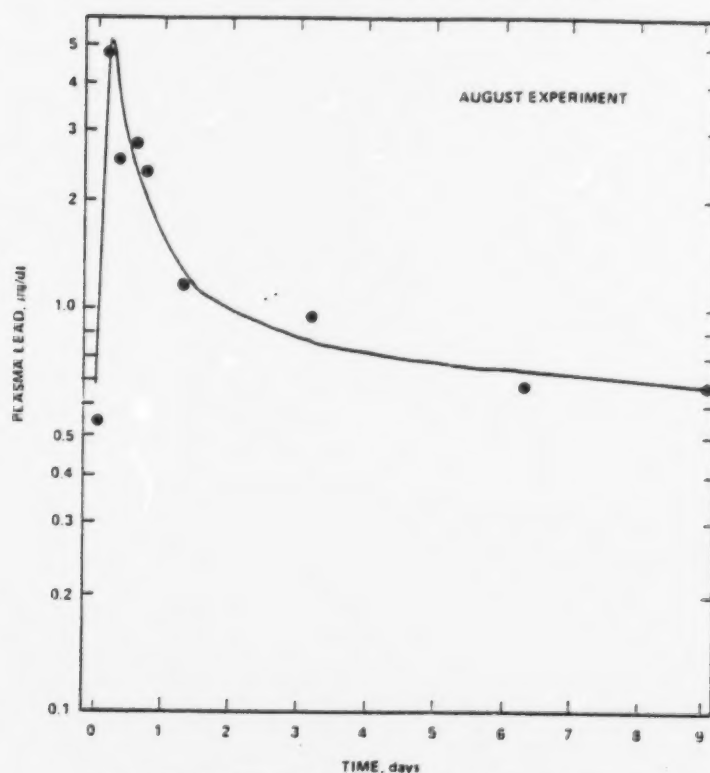


FIG. 5. Observed and predicted plasma lead in August experiment.

these two men was known to have cleaned out bag filters a few hours before being tested." However, even when the largest plasma lead values were deleted from the data set, a significant nonlinear regression remained between plasma and blood lead concentrations. It therefore seems likely that most of the high plasma lead concentrations represent valid near-equilibrium conditions, at least on time scales of a day or so for plasma and erythrocyte kinetics if not on time scales of month so far for whole-body lead kinetics (Rabinowitz *et al.*, 1976, Marcus, 1985b). It seems likely that the nonzero intercept term we recommend using reflects the fact that the plasma lead concentrations in these workers have not yet achieved equilibrium. Recent exposure to lead would elevate the small pool of plasma lead, but only slightly increase the much larger pool of erythrocyte lead.

Multicompartment kinetic models allowed development of explicit equations of plasma lead as a function of blood for various hypothetical nonlinear kinetic mechanisms. If deSilva's data represent valid equilibrium conditions, then the most plausible of the kinetic mechanisms are Models 2 and 4. These models cannot be distinguished using population equilibrium data alone, if an average

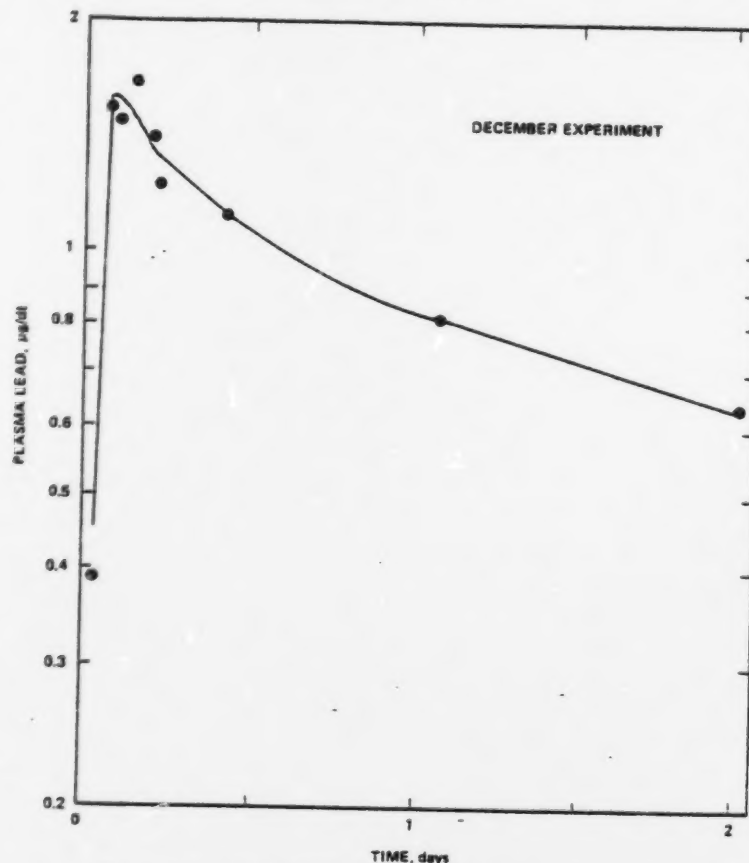


FIG. 6. Observed and predicted plasma lead in December experiment.

hematocrit is assumed. Model 2 corresponds to control of blood lead kinetics by the plasma concentration of lead, either reducing the rate of flow of lead from plasma to erythrocytes in manner much like the Michaelis-Menten theory in enzyme kinetics, or accelerating the rate of flow of lead from erythrocytes to plasma as plasma lead concentration increases. Model 4 corresponds to control of blood lead kinetics by the erythrocyte lead concentration, either reducing the rate of flow from plasma to erythrocyte or increasing the rate of flow from erythrocyte to plasma as the erythrocyte lead concentration approaches its maximum possible value. Other models (e.g., exponential or linear up to a threshold) are possible but cannot presently be justified.

These models were also fitted to a short-term kinetic study in which a high peak blood lead level (78 µg/dl) was achieved. No significant improvement over a linear kinetic model was obtained with any nonlinear model, but between the Models 2 and 4 found best for the population studies. Model 2A (increased plasma

TABLE 7
COMPARISON OF LINEAR KINETIC PARAMETERS^a (PERCENT STANDARD ERROR IN PARENTHESES)

Parameter	August		December	
Fractional transfer rate from/to (day ⁻¹)				
Gut/plasma	64.2	(59.6)	23.8	(26.4)
Plasma/RBC shallow	64.3	(67.9)	634.	(22.6)
Plasma/soft tissue	0.816	(100)	0.822	(97.1)
Plasma/excretion	12.16	(32.8)	12.52	(43.0)
RBC shallow/plasma	2.45	(47.7)	15.2	(29.0)
RBC shallow/RBC deep	1.12	(31.3)	0.604	(49.0)
RBC deep/RBC shallow	0.487	(28.9)	0.401	(65.6)
Fraction absorbed by gut	0.0328	(13.6)	0.0151	(10.9)
Initial pool size (μg Pb)				
Plasma	14.8	(10.0)	10.3	(8.4)
RBC shallow	88.5	(98.6)	84.7	(92.2)
RBC deep	690.6	(17.0)	804.8	(13.4)
Average daily intake (μg Pb/day)	137.8	(62.6)	64.3	(90.9)

^a For experiments reported by deSilva (1981a, b). The assumed rate from soft tissue to plasma is 0.025 day⁻¹. Blood volume = 50 dl, Ht = 0.474.

lead decreases the rate of flow of lead from plasma to erythrocyte by a Michaelis-Menten-like mechanism) proved best for the short-term exposure experiment. This conclusion is, at best, only a suggestion about the mechanisms that need to be explored experimentally.

One biological mechanism that may account for this nonlinearity is the induced formation of lead-binding proteins in erythrocytes (Raghavan *et al.*, 1980). The synthesis of a low-molecular-weight lead-binding protein in the red blood cells (RBC) and the actual physical capture of lead in the RBC does not preclude Model 2A, since the rate-governing kinetic mechanisms such as protein synthesis may be more directly related to the plasma lead concentration than to the erythrocyte lead concentration. We do not presently have a mathematical model for this process.

The amount of lead-binding protein varies greatly among individuals. It is also likely that, due to large changes in lead-binding proteins after exposure to elevated levels of lead, the distribution kinetics of lead may vary significantly with time when even a given individual. The significant differences among all blood kinetic parameters of the subject in Table 7 are otherwise most puzzling. There was little indication of time changes in kinetic parameters of gross lead distribution for any of Rabinowitz's subjects (Marcus, 1985b) except for gut absorption, but Rabinowitz's subjects did not have excessive lead exposure. It is thus likely that their lead-binding protein mass changed only slightly during the experiment.

Difficulties in estimating the nonlinearity parameter $[M]$ for these data led to simulation studies of the estimation procedure in SAAM (Marcus, 1985c). These studies confirmed the insensitivity of the sum of squares criterion. However, "jackknife" estimates of $[M]$ suggest that the data-dependent uncertainty is much smaller than that estimated by the SAAM program (standard error = 0.5 $[M]$).

The present analyses suggest that blood lead may not directly measure the effective toxic burden of lead, particularly at high levels of lead exposure. A very large increase in the toxicologically active plasma concentration of lead may not be matched by a large increase in blood lead. It is thus evident that blood lead alone may not be an adequate criterion for the effects of lead exposure.

We finally caution the reader about the difficulties of plasma lead measurement and about over-reliance on theories which are built on such data.

APPENDIX 1

Linear Compartmental Models for deSilva Study

Let

- $X(1)$ = mass of lead in gut
- $X(2)$ = mass of lead in plasma
- $X(3)$ = mass of lead in shallow erythrocyte pool
- $X(4)$ = mass of lead in deep erythrocyte pool
- $X(5)$ = mass of lead in mobile tissue pool
- $P(1)$ = fraction of ingested lead absorbed through gut.

Then

- (B1) $dX(1)/dt = \text{ingested lead} \cdot P(1) - k(2,1)X(1)$
- (B2) $dX(2)/dt = \text{nonintested lead} + k(2,1)X(1) + K(2,3)X(3) + k(2,5)X(5) - (k(0,2) + k(3,2))X(2)$
- (B3) $dX(3)/dt = k(3,2)X(2) + k(3,4)X(4) - (k(2,3) + k(4,3))X(3)$
- (B4) $dX(4)/dt = k(4,3)X(3) - k(3,4)X(4)$
- (B5) $dX(5)/dt = k(5,2)X(2) - k(2,5)X(5)$

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